CERTIFICATE OF FACSIMILE TRANSMISSION
I HEREBY CERTIFY THAT THIS CORRESPONDENCE WAS TRANSMITTED VIA FACSIMILE TO THE ASSISTANT COMMISSIONER FOR PATENTS, WASHINGTON, D.C., 20231, ATTENTION: EXAMINER A. NELSON, VIA FACSIMILE NUMBER (703) 308-4242, ON OCTOBER 18, 1999.

Catherine D. Brooke
AGENT FOR APPLICANT

October 18, 1999
DATE

Attorney Docket No. 0815

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant:	Jung et al.		
Serial No.:	09/020,716	Group Art Unit:	1649
Filed	2/9/98	Examiner:	A. Nelson
For:	"Alteration of Amino Acid Compositions in Seeds"		

Assistant Commissioner for Patents
Washington, D.C. 20231

DECLARATION OF RUDOLF JUNG UNDER 37 CFR § 1.132

I, Rudolf Jung, declare:

I am a citizen of Germany and a resident of Des Moines, Iowa, United States of America.

I received a degree of Master of Science in Biochemistry from Martin Luther University, Halle, Germany in 1978.

I received a degree of Doctor rerum naturalis in Biochemistry from Martin Luther University, Halle, Germany in 1983.

I presently hold the position of Research Manager, Trait and Technology Development, in the area of improvement of nutritional quality of agricultural seed crops, Pioneer Hi-Bred International, Inc., Johnston, Iowa, 1994-present.

I was a Visiting Scientist in the Biochemistry Department, Purdue University, West Lafayette, Indiana, 1991-1993.

I was a Visiting Scientist in the Agronomy Department, Purdue University, West Lafayette, Indiana, 1990-1991.

I was a Visiting Scientist at Institut fur Genbiologische Forschung, Willmitzer laboratory, Berlin, Germany, 1990.

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I was a Senior Scientist at Zentralinstitut fur Genetik und Kulturpflanzenforschung German Academy of Sciences, Gatersleben, East Germany, 1982-1991 in the area of structure and function of seed storage protein genes in legumes, *Vicia faba*, barley and corn.

I was a Visiting Scientist at Shemyakin Institute of Bioorganic Chemistry, USSR Academy of Sciences, Moscow, Russia, 1987-1988.

I was employed as a Research Fellow at the Institute for Protein Research, USSR Academy of Sciences, Poustchino, Russia, 1978-1981.

The materials and methods for transformation and vector construction are as described in the application.

Appendix A provides data for several independent transformation events for the modified hordothionin (gz::HT12::gz) transformants in maize. I am familiar both with the method employed in obtaining transgenic plants and the methods used in determining the presence of amino acids, especially lysine, cysteine and methionine in seed in Appendix A. Data is expressed in percent dry weight in the meal (e.g. 0.187 = 1.87mg specific amino acid/g of total protein).

The results in Table 1 of Appendix A demonstrate the lysine, cysteine and methionine level of meal from seeds expressing modified hordothionin. These values can be compared to meal from seeds from plants which have not been transformed in Appendix C. Most of the samples of the HT12 transgenics show at least a 10% increase, or at lease a 15% increase or at least a 20% in lysine and in a sulfur containing amino acids over the control. Samples 3-7 and 10-14 show at least a 20% increase in lysine and in a sulfur containing amino acids over the control.

Appendix B provides data for several independent transformation events for the soybean 2S albumin protein (gz::ESA::gz) transformants in maize. I am familiar with the method employed in obtaining transgenic plants and the method used in determining the presence of amino acids, especially lysine, methionine and cysteine in seed in Appendix B.

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The results in Table 1 of Appendix B demonstrate the lysine, methionine and cysteine level of plants expressing modified ESA. These values can be compared to meal from seeds from plants which have not been transformed in Appendix C. Most of the samples of the HT12 transgenics show at least a 10% increase, or at lease a 15% increase or at least a 20% increase in lysine and in a sulfur containing amino acids over the control. Samples ESA 2, 2r, Avg2, 7 and 8 show at least a 20% increase in lysine and in a sulfur containing amino acids over the control.

The results in Table 1 of Appendix C demonstrate the lysine, methionine and cysteine level of control plants. Six independent samples of the same control genotype are shown along with the average % dry weight values for Lysine, Cysteine and Methionine. The average value for each amino acid was compared to the hordothionin and ESA transgenic values in Appendices A and B.

I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that all statements made herein were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title XVIII of the United States Code, and that such willful false statements may jeopardize the validity of this application or any patent issuing hereon.

Rudolf Jung

Dated: 10/18/99

Appendix A

HT-12

HMZ gz::HT12 lines

AA	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Cys	**0.187**	**0.191**	**0.209**	**0.219**	**0.213**	**0.205**	**0.204**	**0.191**	**0.189**	**0.199**	**0.240**	**0.242**	**0.216**	**0.230**
Met	**0.024**	**0.218**	**0.323**	**0.271**	**0.252**	**0.268**	**0.254**	**0.259**	**0.267**	**0.269**	**0.306**	**0.295**	**0.258**	**0.194**
Thr	0.411	0.398	0.379	0.397	0.385	0.383	0.380	0.386	0.413	0.419	0.413	0.424	0.413	0.385
Val	0.522	0.498	0.499	0.508	0.493	0.499	0.495	0.502	0.527	0.515	0.522	0.527	0.512	0.476
Ile	0.394	0.382	0.376	0.376	0.383	0.390	0.376	0.388	0.357	0.370	0.366	0.362	0.370	0.357
Leu	1.399	1.527	1.407	1.395	1.460	1.475	1.464	1.523	1.136	1.310	1.283	1.234	1.332	1.443
His	0.192	0.172	0.173	0.180	0.179	0.181	0.227	0.182	0.235	0.232	0.231	0.230	0.228	0.215
Lys	**0.401**	**0.372**	**0.363**	**0.404**	**0.376**	**0.361**	**0.352**	**0.340**	**0.473**	**0.457**	**0.473**	**0.519**	**0.446**	**0.380**
Arg	0.473	0.396	0.461	0.477	0.526	0.486	0.495	0.468	0.573	0.526	0.505	0.534	0.503	0.364

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Appendix B

ESA AA	ESA1	ESA2	ESA2r	AVG ESA2	ESA3	ESA4	ESA5	ESA6	ESA7	ESA8	ESA9	ESA9r	AVG ESA9
Cys	**0.191**	**0.214**	**0.218**	**0.216**	**0.204**	**0.155**	**0.198**	**0.168**	**0.227**	**0.216**	**0.208**	**0.172**	**0.190**
Met	**0.299**	**0.287**	**0.274**	**0.281**	**0.262**	**0.223**	**0.302**	**0.231**	**0.292**	**0.307**	**0.248**	**0.227**	**0.238**
Thr	0.358	0.375	0.380	**0.378**	0.369	0.355	0.361	0.360	0.384	0.374	0.216	0.359	**0.288**
Val	0.481	0.486	0.485	**0.486**	0.485	0.498	0.492	0.495	0.505	0.496	0.501	0.490	**0.496**
Ile	0.387	0.385	0.380	**0.383**	0.393	0.416	0.387	0.401	0.391	0.383	0.398	0.395	**0.397**
Leu	1.632	1.516	1.459	**1.488**	1.585	1.705	1.587	1.644	1.460	1.496	1.902	1.634	**1.768**
His	0.199	0.215	0.247	**0.231**	0.212	0.180	0.210	0.193	0.223	0.211	0.168	0.208	**0.188**
Lys	**0.207**	**0.273**	**0.268**	**0.271**	**0.250**	**0.215**	**0.221**	**0.218**	**0.301**	**0.269**	**0.160**	**0.214**	**0.187**
Arg	0.408	0.473	0.471	**0.472**	0.450	0.398	0.426	0.396	0.496	0.461	0.463	0.410	**0.437**

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Sample	Lys	Cys	Met
Control 1	0.222		
Control 2	0.221		
Control 3	0.215		
Control 4	0.216	0.151	0.233
Control 5	0.234	0.160	0.228
Control 6	0.223	0.159	0.238
Avg	0.22	0.16	0.23
N (# samples)	6.000	3.000	3.000